[Supplementary Information]

Titanium dioxide nanoparticles-entrapped polyion complex micelles generating singlet oxygen in the cells by ultrasound irradiation for sonodynamic therapy

Atsushi Harada*, Masafumi Ono, Enji Yuba and Kenji Kono

Department of Applied Chemistry, Graduate School of Engineering, Osaka Prefecture University, 1-1 Gakuen-cho, Naka-ku, Sakai, Osaka 599-8531, Japan

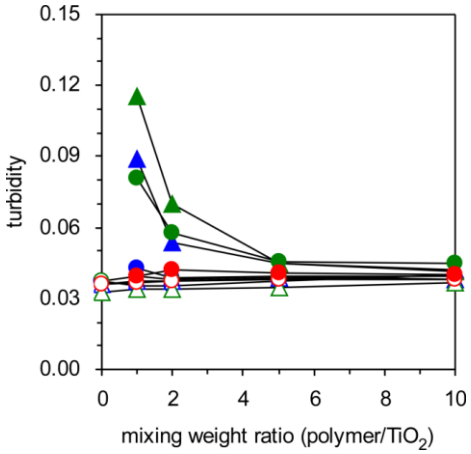

Fig. S1 Change in turbidity with various mixing weight ratios for the mixtures of TiO_2 NPs and PAA-g-PEG before (open symbols) and after (closed symbols) neutralization. The 2k13, 2k26, 5k12, 5k21 and PAA mixtures are represented by blue circles, blue triangles, green circles, green triangles, and red circles, respectively. The pH values before and after neutralization were less than 3 and 7, respectively.

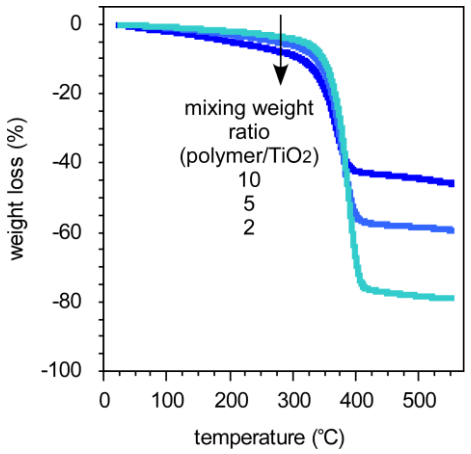

Fig. S2 Typical TG/DTA charts of complexes for the composition determination. The complexes were prepared from mixing weight ratios of 2, 5 and 10 using 5k12. TG/DTA measurements were performed from room temperature to 550°C with a heating rate of 10°C/min. The sample weights were 4.21, 7.01 and 4.38 mg for mixing weight ratios of 2, 5 and 10, respectively.

Table S1 DLS data of TiO_2 NPs-entrapped PIC micelles

PAA-g-PEG	mean diameter (nm)	polydispersity index
2k13	61.0 ± 1.7	0.12 ± 0.02
2k26	86.2 ± 8.5	0.13 ± 0.02
5k12	89.0 ± 6.5	0.12 ± 0.02
5k21	101.8 ± 9.3	0.14 ± 0.02

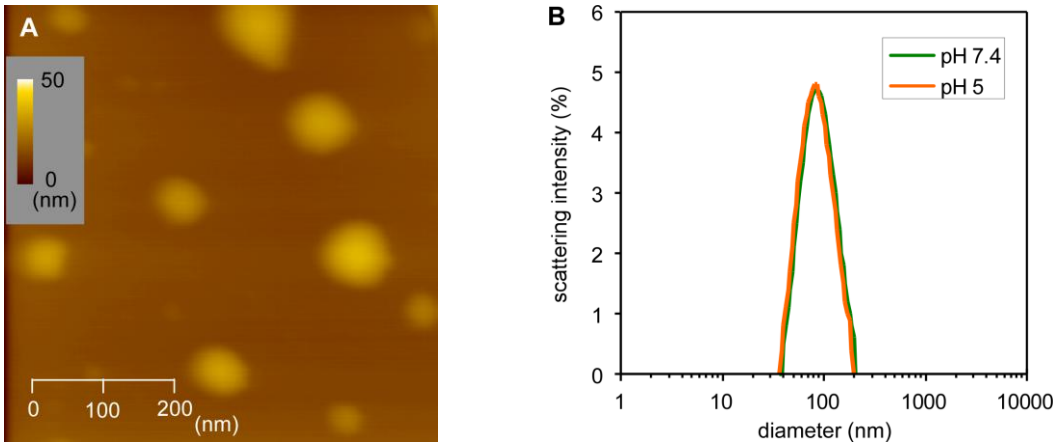

Fig. S3 Morphology observed by atomic force microscopy (A) and size distribution obtained from CONTIN analysis of DLS of TiO_2 NPs-entrapped micelles (2k13 micelles).

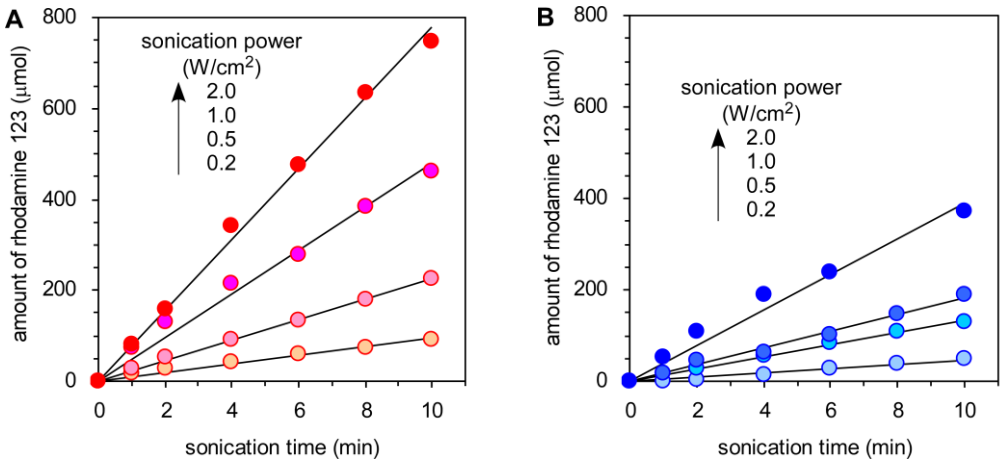

Fig. S4 Evaluation of ROS generation by sonication of TiO_2 NPs-entrapped micelles. Change in the amount of rhodamine 123 produced with (A) and without (B) varying sonication time for 2k26 nanohybrids. The sonication condition was fixed to a frequency of 1 MHz and a duty cycle of 25%. The concentration of DHR-123 and TiO_2 NPs were 10 μM and 45 μg/mL, respectively. Fluorescence measurements were performed at λ_{ex} 480 nm / λ_{em} 530 nm.

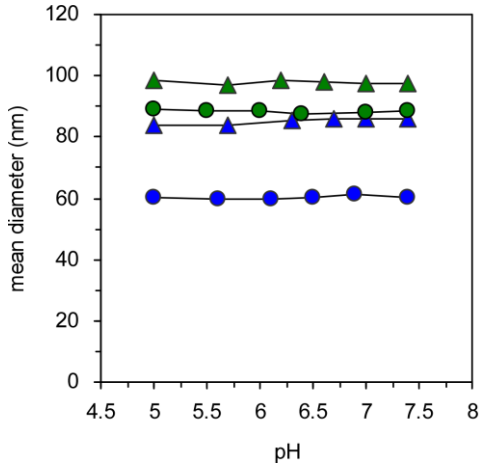

Fig. S5 Change in mean diameter of TiO_2 NPs-entrapped micelles against decreasing pH from 7.4 to 5.0. The 2k13, 2k26, 5k12 and 5k21 micelles are represented by blue circles, blue triangles, green circles, and green triangles respectively. pH was adjusted by adding 0.1M HCl.

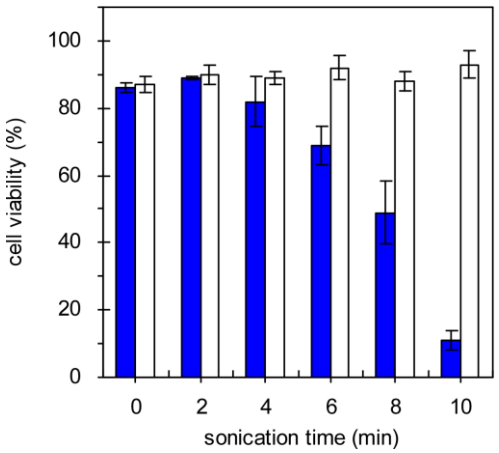

Fig. S6 Effect of sonication time to viability of HeLa cells treated by 2k13 micelles incubated with and without glutathione. The cell viability was determined by MTT assay, and blue and white bars were without and with glutathione. Sonication was performed for varying time (1.0 MHz of frequency, 0.5 W/cm^2 of power and 10% of duty cycle).

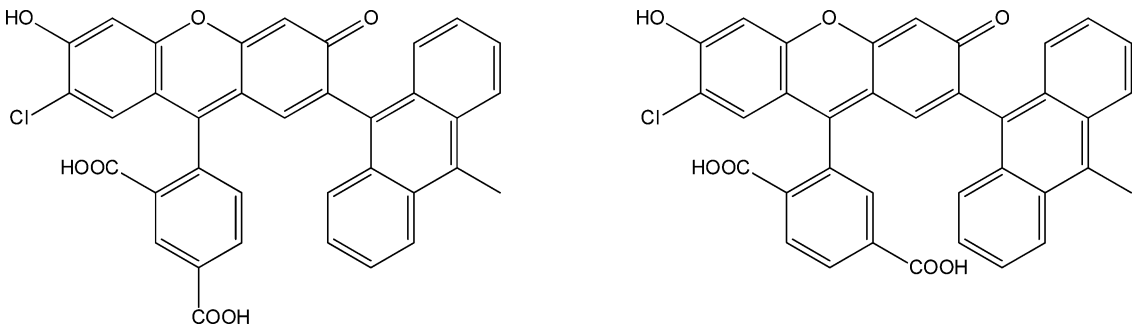

Fig. S7 The expected structure of commercial SOSG. SOSG is likely to a mixture of 4-(7-chloro-6-hydroxy-2-(10-methylanthracen-9-yl)-3-oxo-3H-xanthen-9-yl)isophthalic acid and 2-(7-chloro-6-hydroxy-2-(10-methylanthracen-9-yl)-3-oxo-3H-xanthen-9-yl)terephthalic acid.